

In the Claims

This listing of claims will replace all prior versions and listings of claims in this application.

1 (Currently amended). A recombinant bacterial cell comprising an isolated nucleic acid construct, said cell expressing at least one alkalinizing enzyme and a nickel transporter.

2 (Original). The cell of claim 1, wherein said cell belongs to a bacterial strain that colonizes dental plaque.

3 (Currently amended). The cell of claim 2, wherein said bacterial strain is at least one selected from the group consisting of *Streptococcus mutans*, *Streptococcus sanguinis*, *Streptococcus gordonii*, *Streptococcus parasayzguis*, *Streptococcus vestibularis*, *Streptococcus oralis* ~~of-alis~~, and *Streptococcus mitis*.

4 (Currently amended). The cell of claim 1, wherein said alkalinizing enzyme is an ~~ammonia-producing~~ ammonia-producing enzyme.

5 (Original). The cell of claim 4, wherein said ammonia-producing enzyme is a urease.

6 (Original). The cell of claim 4, wherein said ammonia-producing enzyme is an arginine deiminase.

7 (Original). The cell of claim 4, wherein said ammonia-producing enzyme is an agmatine deiminase.

8 (Currently amended). The cell of claim 1, wherein said construct comprises at least one gene cluster encoding a urease ~~and a nickel transporter~~.

9 (Original). The cell of claim 8, wherein said construct comprises *ureIABCEFGDMQO*.

10 (Original). The cell of claim 1, wherein said construct comprises at least one gene cluster encoding an arginine deiminase system.

11 (Original). The cell of claim 10, wherein said construct comprises *arcABCDTR*.

12 (Original). The cell of claim 1, wherein said construct comprises at least one gene cluster encoding an agmatine deiminase system.

13 (Original). The cell of claim 12, wherein said construct comprises *aguBDAC* and a transcriptional regulator located upstream of the *agu* gene cluster.

14 (Withdrawn). A method for reducing acidification of dental plaque in a subject, comprising the steps of :

providing a subject having at least one tooth with dental plaque disposed thereon, said tooth in a fluid environment, said fluid environment at a first pH;

contacting said tooth with a composition comprising at least one alkalinizing recombinant bacterial cell, said cell colonizing said dental plaque and producing at least one alkali from chemicals in said fluid environment, wherein said alkali raises a pH of said fluid environment to a second pH, said second pH higher than said first pH.

15 (Withdrawn). The method of claim 14, wherein said production of an alkali occurs in the absence of exogenous nickel.

16 (Withdrawn). The method of claim 14, wherein said alkali includes ammonia.

17 (Withdrawn). The method of claim 14, wherein said subject is a human.

18 (Currently amended). A composition comprising at least one recombinant bacterial cell including an isolated nucleic acid construct, said cell expressing at least one alkalizing enzyme and a nickel transporter, in a carrier.

19 (Original). The composition of claim 18, wherein said nucleic acid construct comprises at least one gene cluster encoding a urease enzyme.

20 (Canceled).

21 (Original). The composition of claim 18, wherein said nucleic acid construct comprises at least one gene cluster encoding an arginine deiminase system.

22 (Original). The composition of claim 18, wherein said nucleic acid construct comprises at least one gene cluster encoding an agmatine deiminase system.

23 (Original). The composition of claim 18, wherein said carrier is selected from the group consisting of a chewing gum, a toothpaste, a lozenge, a powder, a gel, an ointment, a cream, a liquid, a mouthwash, a rinse, and a candy.

24 (Withdrawn). An isolated nucleic acid comprising the nucleotide sequence of SEQ ID NO: 1.

25 (Withdrawn). An isolated nucleic acid comprising the nucleotide sequence of SEQ ID NO: 2.

26 (Withdrawn). An isolated nucleic acid comprising the nucleotide sequence of SEQ ID NO: 3.

27 (Withdrawn). An isolated nucleic acid comprising the nucleotide sequence of SEQ ID NO: 4.

28-33 (Cancelled).

34 (Currently amended). A composition comprising:

a host cell; and  
an insertion vector comprising nucleic acid sequences encoding an alkali producing enzyme  
and a nickel transporter; wherein,  
the vector is stably integrated into the host cell genome.

35 (Original). The composition of claim 34, wherein the alkali producing enzyme is urease.

36 (Original). The composition of claim 34, wherein the host cell is selected from cells present in the oral cavity of a subject suffering from dental caries.

37 (Original). The composition of claim 34, wherein the host cell is selected from the genus *Streptococcus*.

38 (Original). The composition of claim 34, wherein the vector targets a *mtl* gene.

39 (Original). The composition of claim 38, wherein the vector further comprises nucleic acid sequences complementary to the *mtl* gene.

40 (Original). The composition of claim 38, wherein the vector invades the *mtl* gene molecule in the region of a target sequence by denaturing the bonds between the complementary target sequences of the double stranded molecule.

41 (Original). The composition of claim 38, wherein the vector is inserted into the *mtl* gene by double-crossover recombination.

42 (Original). The composition of claim 38, wherein the vector is integrated into *mtl* gene in a single copy.

43 (Original). The composition of claim 38, wherein the host cell is selected by its inability to grow on mannitol.

44 (Original). The composition of claim 34, wherein the vector further comprises an entire urease gene cluster.

45 (Original). The composition of claim 44, wherein transcription of the urease gene cluster is under control of a strong promoter.

46 (Original). The composition of claim 44, wherein transcription of the urease gene cluster is under control of a tetracycline resistant promoter isolated from a tetracycline resistant gene.

47 (Original). The composition of claim 44, wherein transcription of the urease gene cluster is under control of a urease cognate promoter.

48 (Original). The composition of claim 34, wherein the host cell further comprises a detectable gene marker.

49 (Original). The composition of claim 48, wherein the detectable gene marker is an antibiotic resistance marker.

50 (Original). The composition of claim 34, wherein the host cell expresses urease enzymes in the absence of any exogenous nickel.

51 (Original). The composition of claim 50, wherein the host cell produces an extracellular alkali base.

52 (Original). The composition of claim 34, wherein the host cell secretes ammonia.

53 (Withdrawn) A method for treating dental caries comprising: a recombinant cell expressing an alkali producing enzyme and a nickel transporter; administering the recombinant host cell to an oral cavity of a subject suffering from or susceptible to dental caries; thereby, neutralizing acidic pH in the subject and treating dental caries.

54 (Withdrawn). The method of claim 53, the alkali produced by the alkali producing enzyme is ammonia.

55 (Withdrawn). The method of claim 53, wherein the recombinant cell is administered in a dose from about  $1 \times 10^4$  up to  $2 \times 10^8$  per mg of bacterial weight.

56 (Withdrawn). The method of claim 53, wherein the recombinant cell produces an alkali, thereby raising acidic pH up to pH 8.

57 (Withdrawn/currently amended). The method of claim 53, wherein the recombinant cell [[is]] belongs to a bacterial strain that colonizes dental plaque.

58 (Withdrawn). The method of claim 57, wherein the bacterial strain is at least one cell selected from the group consisting of *Streptococcus mutans*, *Streptococcus sanguinis*, *Streptococcus gordoyai*, *Streptococcus parasanguis*, *Streptococcus vestibularis*, *Streptococcus oralis*, and *Streptococcus mitis*.

59 (Withdrawn). The method of claim 53, wherein the recombinant cell produces an alkali in the absence of exogenous nickel.